REMARKS

Claims 1-20 have been cancelled without prejudice or disclaimer and redrafted merely for purposes of clarity and to comport with U.S. patent law. Specifically, new claims 21 and 31 are derived from cancelled claim 1. New claims 22 and 32 are derived from cancelled claim 2. New claims 23 and 33 are derived from cancelled claim 5. New claims 24 and 34 are derived from cancelled claim 6. New claims 25 and 35 are derived from cancelled claim 7. New claims 26-27 and 36-37 are derived from cancelled claims 10-11. New claims 28 and 38 are derived from cancelled claim 12. New claims 29-30 and 39-40 are derived from page 9, lines 28-30 of the specification. Applicants reserve the right to pursue in the future any subject matter present in any of the cancelled claims not encompassed by the new claims. No new matter has been added into the claims.

Specification

As suggested by the examiner, applicants have amended the specification to include section headings. The specification was specifically objected to for not having a section entitled, "Brief Description of the Several Views of the Drawings". Applicants have added this section heading on page 6 of the specification after the paragraph ending on line 15 and before the paragraph beginning on line 17. Thus, the objection is no longer applicable and should be withdrawn.

Claim Objections

Claim 3 was objected to for containing confusing grammar. Claim 3 has been cancelled without prejudice of disclaimer and applicants believe that the new claims address the clarity concerns expressed in the office action. Therefore the objection should be withdrawn.

Response to Claim Rejections under 35 U.S.C. § 101

Claims 1-20 have been rejected under 35 U.S.C. § 101, as being directed to non-statutory subject matter by mixing the characteristics of a claim directed to a method with the characteristics of a claim directed to an apparatus. Claims 1-20 have been cancelled without prejudice or disclaimer and rewritten for purposes of clarity as new claims directed to implants (claims 21-30) and new claims directed to methods of using the implant (claims 31-40). Accordingly, the rejection is no longer applicable and should be withdrawn.

Response to Claim Rejections under 35 U.S.C. § 112, Second Paragraph

Claim 1 has been rejected for reciting a use without any active, positive steps delimiting how the use is actually practiced. As noted previously, claims 1-20 have been cancelled without prejudice or disclaimer and new claims directed to implants and method of using the implants have been entered. The claims directed to methods of using the implants properly set forth positive steps for practicing the method.

Claim 1 has been rejected for reciting the term "GSS" without including its name in parentheses at the first occurrence in the claims. The new claims have been drafted to use the term "growth-stimulating substance" instead of its abbreviation "GSS".

Claim 1 has been rejected as being unclear for use of the term "and" together with a second "and/or" in the following phrase, "matrix molecules, growth factors and differentiation factors and/or peptides with growth stimulating properties". The new claims have been amended to clarify this ambiguity.

Accordingly, the rejection has been fully addressed and should be withdrawn.

Response to Claim Rejections under 35 U.S.C. § 112 (Written Description)

Claims 1-20 have been rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the written description requirement with respect to the term "differentiation

factors". According to the rejection, the specification does not identify proteins and peptides that function as differentiation factors. The rejection is improper because differentiation factors are well known in the art and a patent need not teach what is well known in the art. *Ajinomoto Co. v. Archer-Daniels-Midland Co.*, 228 F.3d 1338, 1345 (Fed. Cir. 2000).

Of particular relevance to the instant application is a case recently handed down by the Federal Circuit, *Falkner v. Inglis*, 448 F.3d 1357 (Fed. Cir. 2006). The application in suit was the subject of an interference proceeding and involved vaccines comprising a poxvirus vector having a deleted or inactivated essential gene. Appellants argued that written description was lacking because appellees did not identify in the specification any essential poxvirus genes or the inactivation of any such genes. However, the Federal Circuit agreed with the Board of Patent Appeal and Interferences in determining that the written description requirement was satisfied because the claimed genes and nucleotide sequences were well known in the art. In fact, the court stated that examples are not necessary to support the adequacy of written description and there is no *per se* rule that an adequate written description of an invention that involves a biological macromolecule must contain a recitation of structure. *Id.* at 1366. As one example of the well known nature of differentiation factors, applicants submit the enclosed article, Rose *et al.*, *Bone Grafts and Growth and Differentiation Factors for Regenerative Therapy: A Review*, PRACT. PROCED. AESTHET. DENT. 13(9):725-734 (2001). This is a review article that discusses the state of the art at the time of the invention with respect to differentiation factors and their applicability to regenerative therapy. *See, e.g.*, pages 731-733. Accordingly, the rejection is improper and should be withdrawn.

Response to Claim Rejections under 35 U.S.C. § 102(b)

Claims 1-4, 6-9, 11, and 14-17 have been rejected under 35 U.S.C. § 102(b) as being anticipated by Wikesjo *et al.*, *Augmentation of Alveolar Bone and Dental Implant Osseointegration: Clinical Implications of Studies with rhBMP-2*, J. BONE AND JOINT SURGERY, 83:S1-136 through S1-145 (April 2001) ("Wikesjo *et al.*"). The rejection is improper because Wikesjo *et al.* does not describe every element of the claims. A claim is anticipated only if each and every element as set forth in the claim is found, either expressly or inherently described, in a

single prior art reference." *Verdegaal Bros. v. Union Oil Co. of California*, 814 F.2d 628, 631 (Fed. Cir. 1987); § MPEP 2131.

The instant claims are drawn to an implant and methods for building up bone based lateral support, wherein bioactive or osteoinductive material is in or on the implant and the implant is arranged so that it forms spaces together with the soft tissue and/or a unit and the upper or lateral surface(s) of the jaw bone. Wikesjo *et al.*, however, evaluates surgical implantation of rhBMP-2 delivered by carriers such a bovine collagen sponge and decalcified bone matrix used as onlays or inlays in conjunction with implants, *i.e.*, the rhBMP-2 is not part of the implant *per se*. Wikesjo *et al.*, p. S1-140. Also, the implants of Wikesjo *et al.* do not form spaces together with the soft tissue and/or a unit and the upper or lateral surface(s) of the jaw bone. One end of the implant is anchored into the jawbone and the other end of the implant extends into a surgically reduced edentulous mandibular ridge. *Id.* Thus, Wikesjo *et al.* cannot anticipate the instant claims.

Response to Claim Rejections under 35 U.S.C. § 103(a)

Claims 1, 5-8, 10, 12-13 and 18-20 have been rejected under 35 U.S.C. § 103(a) as being obvious over Wikesjo *et al.* and Pirhonen *et al.*, U.S. Patent Application Publication No. 2003/0105530 (Pirhonen *et al.*). The rejection is improper because (1) all the elements of the claims are not accounted for in the cited references and, (2) Wikesjo *et al.* teaches away from what is claimed.

First, the references do not account for every element of the claims. To establish *prima facie* obviousness, all the elements of the claims must be found in the prior art. *In re Vaeck*, 947 F.2d 488 (Fed. Cir. 1991); *In re Royka*, 490 F.2d 981 (CCPA 1974). With respect to all the claims, the combination of references do not account for an implant having bioactive or osteoinductive material in or on the implant and do not account for spaces formed between the implant and soft tissue and/or a unit and the upper or lateral surface(s) of the jaw bone.

Additionally, with respect to claims 22 and 32, the combination of references does not account for an implant being offset so that side surfaces or outer thread surfaces of the implant are exposed.

With respect to claims 23 and 33, the combination of references does not account for an implant having an exposed surface presenting a higher concentration of bioactive or osteoinducive material than non-exposed surfaces of the implant.

With respect to claims 26-27 and 36-37, the combination of references does not account for an exposed surface of 20-180° in the circumferential direction and the height direction of the implant.

With respect to claims 28 and 38, the combination of references does not account for a bioactive or osteoinductive coating on the surface of the unit exposed toward the implant.

With respect to claims 30 and 40, the combination of references does not account for an implant coated with an oxide layer having pores in which the bioactive or osteoinductive material is stored.

Second, Wikesjo *et al.* actually teaches away from proceeding as applicants have done by suggesting that first rhBMP-2 should be applied to the jawbone to induce bone growth and later anchoring the implant into the new bone growth instead of using rhBMP-2 as part of the implant to induce bone growth around the implant. Where the teaching of a reference discourages persons skilled in the art from doing what applicants claim, the reference established "the very antithesis of obviousness." *In re Buehler* 185 USPQ 781 (CCPA 1975); *In re Rosenberger and Brandt*, 156 USPQ 24, 26 (CCPA 1967); *See also* MPEP(X)(D)(1)-(3).

In an experiment by Caplanis *et al.* described on page S1-140, the authors evaluated the surgical implantation of decalcified bone matrix containing rhBMP-2 in conjuction with guided bone regeneration using two implants (*i.e.*, the decalcified bone matrix and the implant were implanted at the same time). This procedure resulted in only limited bone formation along the exposed implant surfaces. *See, e.g.*, p. S1-140, bottom of the second column. However, in a

subsequent experiment by Sigurdsson *et al.*, described on page S1-141, second column, the authors first applied an onlay of decalcified bone matrix containing rhBMP-2 to the jawbone (to induce bone growth) and then placed the implants into the new bone growth 8 and 16 weeks later. The authors reported that approximately 90% of the bone anchoring surface of the implants was invested in newly induced bone suggesting that a better result is obtained by first inducing bone growth with rhBMP-2 and then later using the new bone growth to anchor an implant. Applicants, however, have proceeded against this suggestion and claimed an implant and method of using the implant wherein the bioactive or osteoinductive material is located in or on the implant and therefore incorporated together (at the same time) into the jawbone before new bone is formed.

In sum, the rejection is improper and should be withdrawn because all the elements of the claims are not accounted for by the combination of references and because Wikesjo *et al.* teaches away from proceeding as applicants have done.

In view of the above, consideration and allowance are respectfully solicited.

In the event the Examiner believes an interview might serve in any way to advance the prosecution of this application, the undersigned is available at the telephone number noted below.

The Office is authorized to charge any necessary fees to Deposit Account No. 22-0185.

Enclosed is the fee and petition for a three-month extension of time. Applicants believe no additional fees are due with this response. However, if a fee is due, please charge our Deposit Account No. 22-0185, under Order No. 21547-00304-US1 from which the undersigned is authorized to draw.

Dated: August 5, 2008

Respectfully submitted,

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Bone Grafts and Growth and Differentiation Factors for Regenerative Therapy: A Review

Louis F. Rose, DDS, MD*
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Guided bone regeneration, tissue grafts, regenerative barrier membranes, and bone substitute materials have been used to restore inadequate hard and soft tissue structures to make them conducive to proper implant placement. Polypeptide growth and development factors (GDFs) have successfully been applied exogenously to periodontal defects to attract preosteoblasts to the site and accelerate their proliferation to stimulate angiogenesis. This article provides an overview of current modalities for restoring lost bone and soft tissue during the treatment of periodontal disease.

Key Words: growth factors, regeneration, osteoinduction, osteoconduction, graft

Conventional periodontal treatments, such as root planing, gingival curettage, and scaling, are highly effective at repairing disease-related defects and halting the progression of periodontitis. While these are important steps, researchers are still challenged to develop more effective techniques that predictably promote the body's natural ability to regenerate its lost periodontal tissues — particularly alveolar bone.

In addition, the focus of implant restoration treatment for periodontitis patients has expanded during the past decade to include both function (adequate osseointegration) and aesthetics. The goal now is to create a restoration that rivals the natural dentition in terms of performance, comfort, and appearance. This requires an optimal hard and soft tissue complex that harmonizes with the patient's natural teeth. It also requires a team effort by all clinicians involved as well as direction from the patient in terms of what constitutes an acceptable aesthetic result. Various therapeutic modalities are currently used to restore inadequate bone and soft tissue to make them conducive to proper implant placement. These include guided bone regeneration, bone and soft tissue grafts, the use of regenerative barrier membranes, and placement of bone substitute materials (Figure 1).

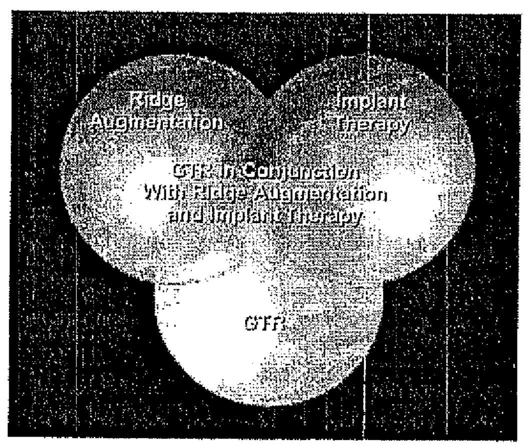

Figure 1. Diagram demonstrates that integration of therapy is essential to establish aesthetic and functional success.

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In recent years, clinicians have also begun to learn more about how periodontal regeneration works on a cellular and molecular level. This is a key step to developing new strategies and materials for promoting predictable periodontal regeneration. The most promising new technique currently being investigated is the application of polypeptide growth and development factors (GDFs), natural proteins in the body that regulate the wound and tissue regeneration. Based on several in vivo studies and early human clinical trials, investigators are moving closer to identifying specific combinations of GDFs that can be applied exogenously to a periodontal defect for attracting preosteoblasts to the site, accelerating their proliferation, and stimulating angiogenesis. This article provides an overview of current treatments for restoring lost bone and soft tissue in patients with periodontal disease and information regarding the latest research on GDFs.

Ridge Augmentation

Alveolar bone grafting is the most common form of regeneration therapy today, and it is generally essential for restoring all types of periodontal supporting tissue. To date, histological evidence in humans indicates that bone grafting is the only treatment that results in the regeneration of bone, cementum, and a functionally oriented, new periodontal ligament (PDL) coronal to the base of a previous osseous defect (Figures 2 and 3).[1,2] It has also been shown to produce a successful clinical result

Figure 2. Case 1. An alveolar ridge of insufficient width requires ridge expansion to accommodate implant placement.

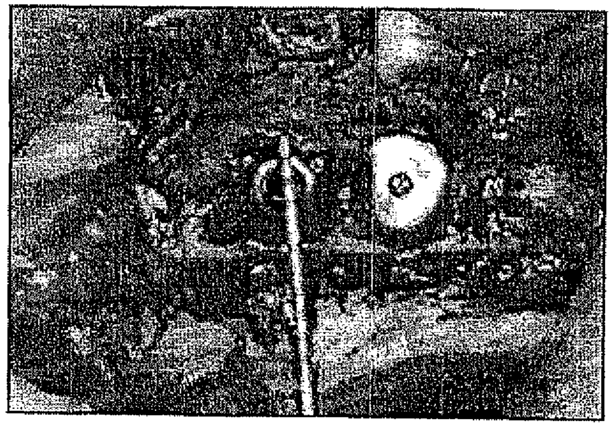

Figure 3. A chisel is used to expand the buccal and palatal bone to a minimum width of 5 mm to 6 mm to accommodate the implants.

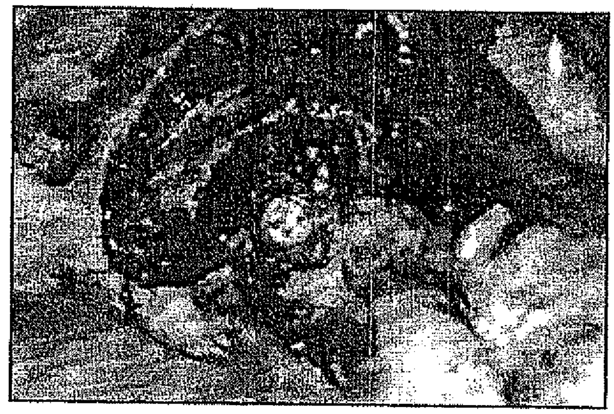

Figure 4. Case 2. Clinical view of small alveolar ridge defect with a titanium tenting screw that will help to maintain space necessary for bone regeneration.

that lasts more than 20 years when patients effectively control plaque through proper oral hygiene and regular professional periodontal maintenance visits.[3]

Types and Application of Graft Materials

Several types of bone and bone substitutes have been studied over the years, and periodontists continue to search for ideal materials. The type of graft material used in a particular case is frequently dictated by the amount of bone that must be replaced. The two most common types of graft material used in periodontology are autogenous and allogenic. Autogenous bone is considered the "gold standard," particularly for treating large deformities.

For implant restorations, sinus augmentation may be necessary in addition to ridge augmentation. This allows

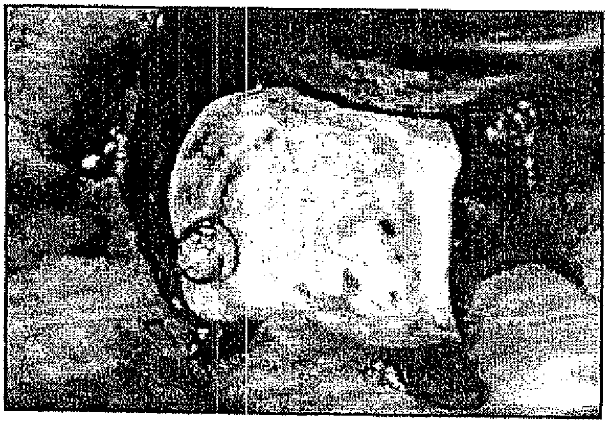

Figure 5. Autogenous bone is harvested from the tuberosity and placed into the defect. A barrier membrane was replaced over the graft and stabilized with a titanium screw.

Figure 6. Placement of three implants within the reconstructed alveolar ridge 8 months following bone regeneration.

proper placement of a sufficient number of implants as well as appropriate sizes in the maxillary posterior areas. Sinus augmentation is indicated in particular when the interocclusal distance cannot accommodate an onlay graft.[3] Thus, a combination of sinus and ridge augmentation is often used to achieve an optimal hard tissue environment for implant placement.

Autogenous Grafts

Autogenous grafts, which are harvested from one part of the patient's body for transplantation to another, are considered optimal due to their superior retention of cell viability. These grafts contain live osteoblasts and osteoprogenitor stem cells, which proliferate and bridge the gap between the graft and the recipient bone (Figures 4 through 8).[3] The area of new bone interdigitation and the quantity of donor bone that is resorbed are greater with cancellous bone grafts than cortical grafts.

Case reports show that the average bone fill when using autogenous grafts is approximately 3.5 mm for many intraosseous defects, depending on their initial size. Histological evaluations suggest that at least partial periodontal regeneration occurs following these types of grafting procedures.[4] In addition, autogenous grafts avoid the potential problems of histocompatibility differences and the risk of disease transfer that other allograft and xenograft types pose.[5]

Autogenous bone should be used whenever possible, and it can be harvested either from intraoral or extraoral sites. Intraoral cancellous bone and marrow grafts are usually obtained from the maxillary tuberosity or a healing extraction site.[6] Extraoral sites include the iliac crests, ribs, cranium, and tibial metaphyses (Figures 9 through 16). While patients frequently consent to autogenous bone grafts from intraoral sites, sufficient amounts of graft material are not always available. In addition, many patients refuse or cannot afford to be hospitalized for extraoral graft procurement. These are common obstacles to the use of autogenous grafts.

Allografts

Due to these limitations, allografts were developed as an alternative. Allografts are harvested from one human for transplantation to another. These grafts are typically

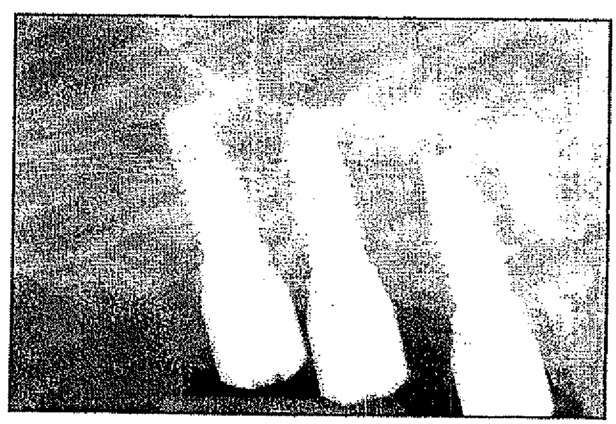

Figure 7. Six-month postoperative radiograph demonstrates successful placement of implants into the alveolar ridge.

freeze-dried and treated to prevent disease transmission. Various types of allografts are available from commercial tissue banks, including frozen iliac cancellous bone and marrow, freeze-dried bone, and demineralized freeze-dried bone allograft (DFDBA).

There is some debate about how allografts heal. Some authors believe they heal by osteoinduction. Over time, the allograft is resorbed by the natural intraoral bone, and this regenerative process is thought to be induced by bone morphogenic protein (BMP) and perhaps other GDFs released from the allograft.[7,8] Demineralized freeze-dried bone grafts appear to heal by osteoinduction.[9] Many authors, however, are not convinced that osteoinduction actually occurs, even though it appears that freezing or freeze-drying graft material does not destroy its cellular activity. Some contend that allografts may heal by osteoconduction, a process in which the graft does not activate bone growth, but instead acts as a scaffold onto and within which the patient's own natural bone grows.[10]

Frozen iliac allografts are rarely used today due to the need for extensive cross-matching to minimize the risk of graft rejection or disease transmission. Mineralized freeze-dried bone (FDBA) is still used, but a recent large-scale research review showed that FDBA mixed with autogenous bone is more effective at increasing bone fill than FDBA alone.[10]

Demineralized freeze-dried allograft is the most widely used allograft material in periodontics today due to its availability, safety, and its purported osteoinductive and osteoconductive properties.[11] In the 1960s and 1970s, Urist et al showed that demineralizing and freeze-drying graft material dramatically enhanced its osteogenic potential. Removing the bone mineral appears to be a crucial factor. This process exposes BMPs or other GDFs in the graft material that stimulate the formation of new bone by osteoinduction.[12] Human clinical studies have shown DFDBA grafts result in 2.5 mm to 3 mm of bone fill (Figures 17 through 20).[10]

Debating DFDBA Effectiveness

Although human histological evidence indicates that DFDBA can promote the formation of new attachment apparatus on root surfaces,[2] some researchers and clinicians have recently begun to question its usefulness.

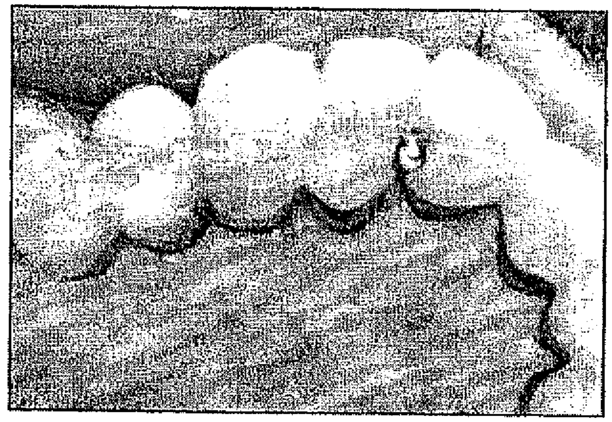

Figure 8. Occlusal view of the final implant-supported restoration 36 months postoperatively.

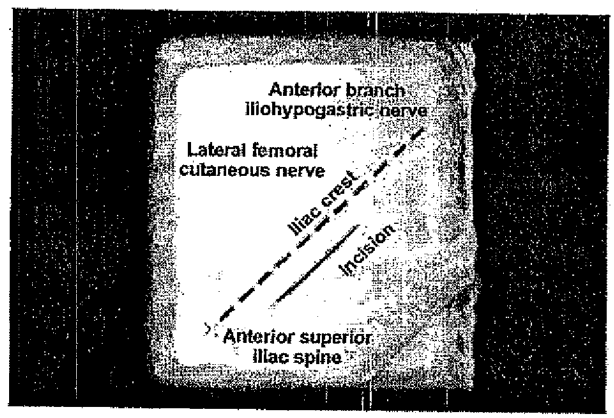

Figure 9. Case 3. Illustration demonstrates the anatomy and incision lines necessary to harvest bone from the iliac crest.

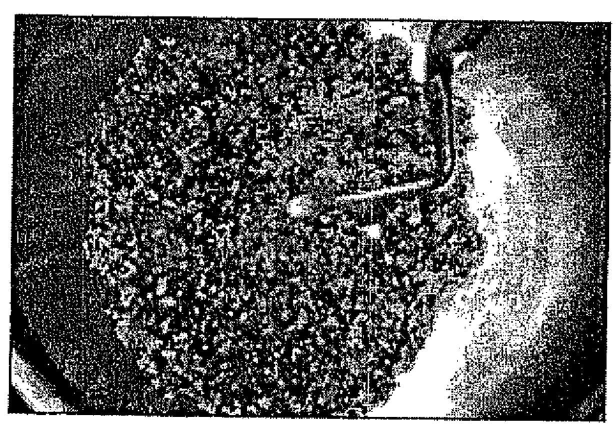

Figure 10. Once particulate cancellous bone has been harvested from the iliac crest, it is prepared for placement in the maxillary sinus.

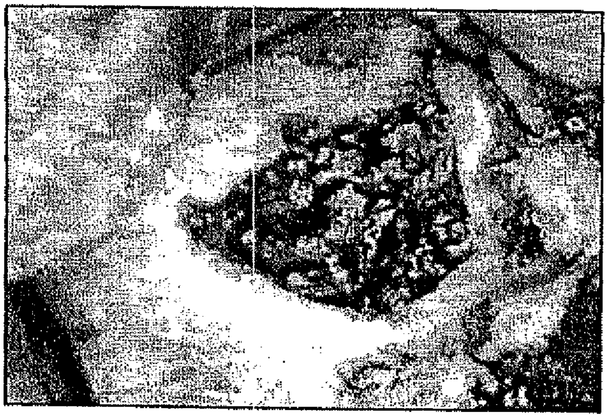

Figure 11. Maxillary sinus after completion of bone fill with particular cancellous graft.

Figure 12. Cortico-cancellous veneer grafts are utilized to increase the width of the maxillary alveolar ridge.

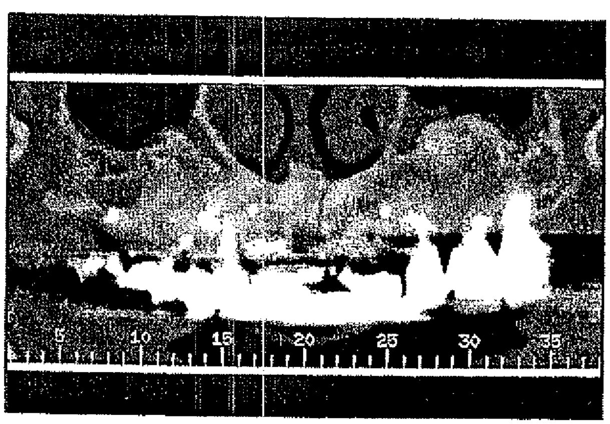

Figure 13. Eight-month postoperative panoramic radiograph of connective tissue healing.

Contrary to studies that have found good regenerative results with DFDBA,[2,13-15] other studies show less predictable and even disappointing results with this material.[16-18] One likely explanation is that DFDBA material may vary from graft to graft. Commercial bone banks do not verify the specific amount of BMPs or any level of inductive capacity in any graft material they sell. Therefore, graft quality is not standardized. This was demonstrated in one recent study by Schwartz et al,[19] who obtained DFDBA from six bone banks and from different lots at some of the same banks. They evaluated the ability of the material to induce bone formation in an in vivo model with 49 mice over a 2-month period. These investigators found that the different DFDBA preparations varied significantly in their particle size and inductive properties (although particle size did not appear to have any bearing on inductive capacity in their study). In addition, the batches varied considerably in their abilities to promote bone and cartilage formation. This notable inconsistency was even evident among two DFDBA batches from the same bank — one showed some osteoinductive activity, and the other showed none whatsoever. These findings are supported by other studies.[20,21]

Delaying the procurement of bone from a donor, improper storage conditions, or other processing factors may play a significant role in the bioactivity of the final DFDBA preparation that is forwarded to the clinician's office. The significant effects of these factors were demonstrated in studies that compared laboratory and commercially prepared extracts.[22,23] Age, gender, and medical status of deceased donors may also affect osteogenic activity in the grafts taken from them. Some authors suggest that more predictable results might be achieved with DFDBA grafts if bone banks standardized graft material by instituting strict standards on the sources and time frames associated with procuring grafts and by developing a way to quickly test the inductive capacity of any graft materials they supply.[19,24] Studies have determined that the minimum effective amount of BMP necessary to affect bone growth is approximately 2 μg/40 mg weight of explant; the optimum amount is approximately 10 μg/40 mg.[25]

Another issue concerns the fate of DFDBA material placed in periodontal defects over time. Some studies

Figure 14. Clinical view demonstrates healing of the veneer graft 9 months postoperatively.

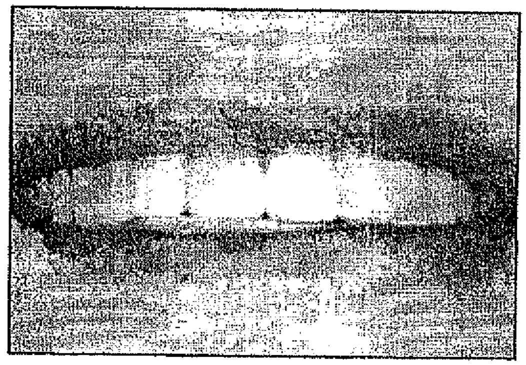

Figure 15. Facial view of the patient at relaxed smile evidences the final implant-supported restoration.

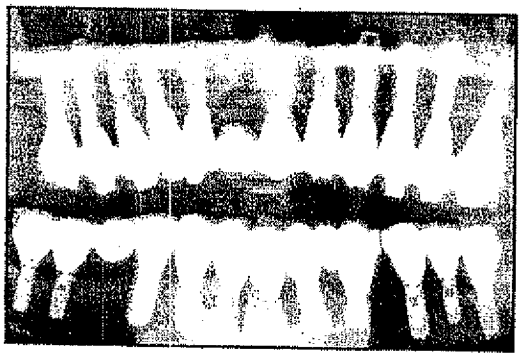

Figure 16. Postoperative radiograph 6 years following placement of the implant supported maxillary and mandibular fixed restoration.

Figure 17. Case 4. Alveolar ridge immediately after extraction of natural teeth. Note the lack of facial bone.

indicate that residual DFDBA particles remain in the site for longer than a year and act as bone matrix.[12,26] It is unclear how these leftover particles might affect the regenerative response. Becker et al noted that retained dead bone chips appeared to delay normal bone formation in sockets and to weaken host bone.[17] In a histological analysis, however, Reynolds and Bowers found that grafted defects harboring residual DFDBA particles had significantly more new attachment formation than sites without residual particles and that the regenerated tissues in both cases had similar characteristics.[27]

Alloplasts

Alloplasts, or bone substitutes, have also been developed for ridge augmentation (Figures 21 through 24). These materials include biocompatible porous and nonporous hydroxyapatite, bioactive glass, beta tricalcium phosphate, and HTR polymer.[28] Clinically, alloplasts act as biological fillers and have been shown to improve probing depth and clinical attachment. They do not revascularize by microanastomosis, but rather by capillary sprouts that invade the implant from the host bed during resorption of the old matrix. Alloplasts heal by osteoconduction, and some are quite resistant to resorption.[9] Histologically, however, connective tissue tends to encapsulate bone substitutes, and they induce minuscule bone fill and little, if any, periodontal regeneration.[4,29] Currently, these may be useful as allograft extenders.

Soft Tissue Reconstruction

To achieve optimal aesthetic results in restoring periodontal defects, the clinician must attend to the soft tissues as well as bone. This is particularly important when implants are placed. Soft tissue reconstruction requires the clinician to accurately delineate the specific defect and to recognize the quantity and quality criteria necessary to restore the tissue anatomy. Surgical procedures that can be used to restore soft tissues include various

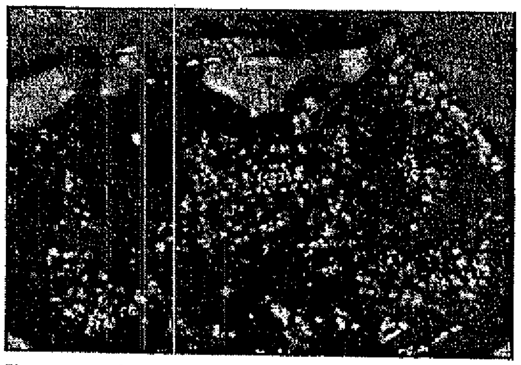

Figure 18. Palatal view exhibits placement of freeze-dried decalcified bone in the bone deformities.

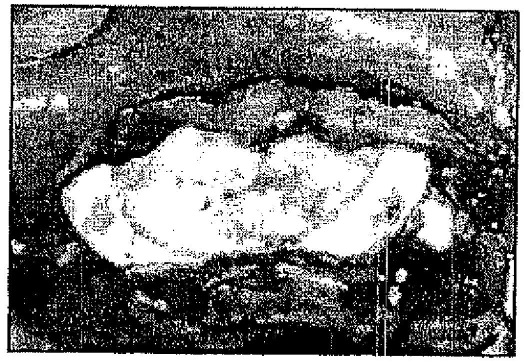

Figure 19. Placement of e-PTFE membrane over the allograft to protect site and promote the healing process.

Figure 20. Eight-month postoperative result demonstrates bone regeneration. Note enhanced thickness of ridge.

Figure 21. Case 5. Clinical view of small postextraction alveolar ridge defect.

flap designs, onlay grafts, subepithelial connective grafts, and tissue expansion. These treatments are also used to augment the interproximal papillae and to cover reconstructed hard tissues with primary tension-free closure during healing. In implant cases, they can establish the foundation necessary to enhance implant position, size, angulation, and emergence profile.

Restorative procedures (eg, provisional restorations and appropriate abutments) and orthodontic procedures can also be useful in establishing soft tissue aesthetics. Orthodontics can help to prevent a ridge deformity after a hopeless tooth is extracted. For instance, Salama et al reported the use of forced eruption, a nonsurgical approach, for regenerating hard and soft tissues around a hopeless tooth.[29] Periodontal orthodontics are used to increase the vertical osseous dimension and to preserve the papilla; restorative orthodontics optimizes the implant site by manipulating supragingival restorative space.

Growth and Differentiation Factors

Perhaps the most promising research currently underway involves the use of inductive proteins, known collectively as polypeptide GDFs. These molecules are key regulators of cellular wound repair for nearly all tissues, including the periodontium.[30-32] When a wound occurs, several growth factors are released from within and around the wound site. They work together to regulate cell activity and to repair and regenerate the various types of tissue. How well these GDFs are expressed during the progression of periodontal disease and associated bone and soft tissue injuries is not known, but this may profoundly influence the repair or regenerative process. Therefore, the goal of administering GDFs to treat chronic periodontitis would be to enhance a normal wound healing process that may otherwise be insufficient to completely regenerate the entire periodontal attachment apparatus.[33] This is the subject of current experimental research, and human trials have just begun.

During the past decade, several growth factors have been identified and characterized. Growth and differentiation factors currently believed to contribute to periodontal regeneration include platelet-derived growth factor (PDGF), transforming growth factor-beta (TGF-β), insulin-like growth factor (IGF), fibroblast growth factor (FGF), and BMPs.[34] These periodontal growth factors have some common features, including that they bind to specific receptors on the target cell surface, they primarily act locally, and they stimulate a variety of cell activities.[35]

One important characteristic researchers have identified is that growth factors are cell specific.[36] In other words, particular growth factors work only to stimulate specific cell types. Clearly, a combination of GDFs, including those that stimulate wound closure and the formation of hard and soft tissues, is necessary.

Considered one of the principal wound healing hormones, PDGF has been the most studied growth factor. Platelet-derived growth factor is a potent mitogen and chemotactic factor for cells of mesenchymal origin, which include PDL cells and osteoblasts. There may be several cellular sources (eg, platelets, activated macrophages, bone matrix) of PDGF at a wound site, and PDGF has been shown to stimulate the proliferation of osteoblasts and periodontal ligament cells that have both fibroblastic and osteoblastic characteristics. When applied to root surfaces of teeth with surgically created defects in six dogs, PDGF enhanced fibroblast proliferation in the early healing stages.[34]

Thus far, the results have been promising in animal studies and some preliminary human studies involving the use of GDFs. Most of the studies have examined the effect of PDGF, either alone or combined with other growth factors, to promote periodontal repair. In the recently reported first human trial, 38 patients with moderate-to-severe periodontitis were treated with either a) 150 μg/mL each of PDGF-BB and IGF-1 in a methylcellulose vehicle, b) vehicle alone, or c) surgery alone. Those treated with the PDGF/IGF combination experienced a 43% bone fill, while the control groups had an average 18.5% fill.[36] Several previous animal studies on PDGF/IGF combinations have also shown encouraging results in the promotion of new bone, cementum, and periodontal ligament.[37] Some have demonstrated that

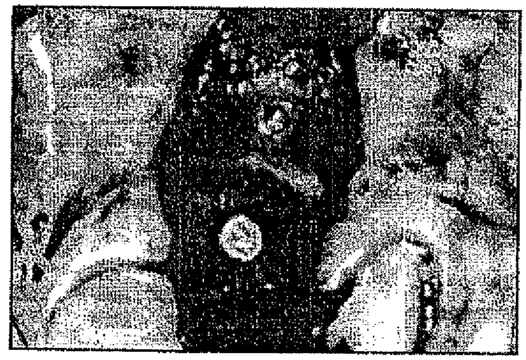

Figure 22. Two titanium tenting screws are utilized to assist in space maintenance.

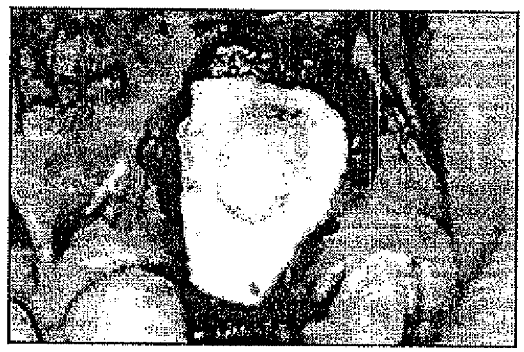

Figure 23. A bioactive glass alloplast is used in conjunction with an e-PTFE membrane to augment the alveolar ridge.

this combination of growth factors also enhances bone growth around implants placed into fresh extraction sockets.[38] The mechanism by which this growth factor combination improves periodontal regeneration must still be proven in vivo, as do precise amounts of the combination that would be optimally useful.

Latent TGF-β is primarily stored in bone matrix, and while the mechanism that activates it is unclear, some speculate that a low pH during osteoclastic bone resorption may be responsible.[34] Almost every cell type can be stimulated by at least one of the five types of gene-encoded TGF-β molecules, which is a weak mitogen for osteoblasts. Also inducing chemotaxis and stimulating collagen type-1 synthesis, TGF-β has been shown to have a positive preferential mitogenic effect on PDL cells over gingival fibroblasts. It also inhibits reepithelialization. Recent in vitro studies suggest that the combination of TGF-β and PDGF may selectively stimulate PDL cells more

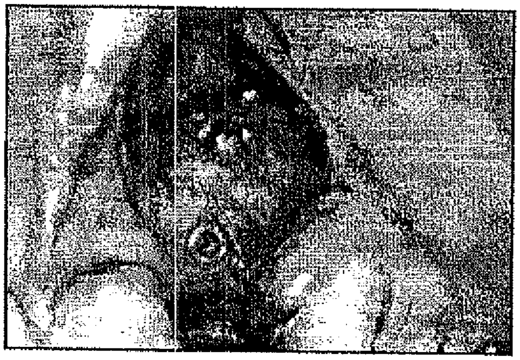

Figure 24. At 9 months postsurgery, the alveolar ridge is reexamined. Note the improved bone volume.

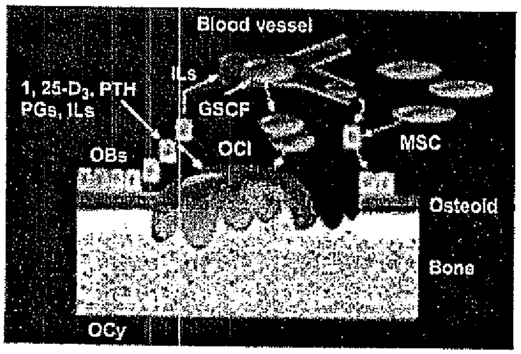

Figure 25. Illustration depicts the biological processes at the cellular and tissue levels during bone formation. During the process of remodeling, osteoblasts (OB) and osteoclasts engage in a dynamic interaction.

than gingival fibroblasts, thus enhancing periodontal regeneration.[39,40] Further in vivo studies are necessary to confirm this.

There are two types of IGFs, which function similarly but are independently regulated. They are produced primarily by the liver and circulate in the vascular system. Insulin-like growth factor-1 appears to stimulate bone formation by increasing cellular proliferation and bone matrix production (Figure 25). As mentioned, however, in vivo animal studies and the recent clinical trial have shown that it appears to work better in combination with PDGF than on its own. The IGF-1 has also been shown to stimulate mitogenic activity and chemotaxis in PDL cells.

Fibroblast growth factors are mitogenic and chemotactic for fibroblasts, chondrocytes, osteoblasts, and endothelial cells. These are stored in bone matrix as well. Although FGF increases the number of osteoblasts capable of regenerating bone, it decreases the amount of matrix each cell produces. The net effect, however, appears to be enhanced bone formation. The FGFs are unique in that they are potent angiogenic factors, stimulating the formation of blood vessels that are critical to wound healing and granulation tissue formation. Again, additional in vivo studies are necessary to determine a treatment benefit.

Bone morphogenic proteins are unique in that they have osteoinductive properties. The BMPs, which are a member of the transforming growth factor (TGF) gene superfamily, are believed to play a significant role in recruiting osteoprogenitor cells to sites of bone formation.[41] They are able to stimulate the differentiation and proliferation of mesenchymal stem cells into chondroprogenitor and osteoprogenitor cells. Various in vivo studies in animals have demonstrated that BMPs induce significantly more cementum, periodontal ligament, and bone regeneration in surgically created defects than do untreated controls.

The use of osteogenin, one of several different forms of BMP, has shown promise in studies with dental implants. In association with a bone-driven matrix, osteogenin rapidly initiates bone formation.[42] Investigators found that combining osteogenin with DFDBA significantly enhanced regeneration of a new attachment apparatus (1.92 mm) compared with DFDBA alone (1.31 mm) when submerged, although the improvement was not statistically significant in a nonsubmerged environment.[15] Wang et al found similar results noting that BMPs induced earlier bone apposition around implants.[36] By accelerating osseointegration, BMPs might allow earlier loading of implants with permanent restorations.

Conclusion

The future clinical application of growth factors in regeneration therapy is promising. Indeed, some researchers have noted that they may eventually replace autogenous or allogenic grafts for promoting osteoinduction in defect sites.[43,44] First, however, researchers will need to determine the ideal combination of factors for regeneration, the best delivery system, and optimum doses. It is also important to note that growth factors may need to be delivered in a precise sequence during healing to afford any regeneration benefit.

Acknowledgment

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Continuing Education (CE) Exercise No. 29

29
CONTINUING EDUCATION

To submit your CE Exercise answers, please use the answer sheet found within the CE Editorial Section of this issue and complete as follows: 1) Identify the article; 2) Place an X in the appropriate box for each question of each exercise; 3) Clip answer sheet from the page and mail it to the CE Department at Montage Media Corporation. For further instructions, please refer to the CE Editorial Section.

The 10 multiple-choice questions for this Continuing Education (CE) exercise are based on the article "Bone grafts and growth and differentiation factors for regenerative therapy: A review" by Louis F. Rose, DDS, MD, and Edwin Rosenberg, DMD, BDS, HdipDent. This article is on Pages 725-734.

Learning Objectives:

This article provides an overview of current modalities for restoring lost bone and soft tissue during the treatment of periodontal disease. Upon reading this article and completing this exercise, the reader should:

- Understand the role of growth and differentiation factors in the success of tissue regeneration.
- Be aware of the various types of bone and bone substitutes available as graft materials.

1. Autogenous grafts:
   a. Provide superior retention of cell viability.
   b. Contain live osteoblasts and osteoprogenitor stem cells.
   c. Are harvested from one part of the patient's body for transplantation to another.
   d. All of the above.

2. Polypeptide growth and development factors:
   a. Should not be applied exogenously to periodontal defects.
   b. Attract preosteoblasts to treatment sites and decelerate their proliferation.
   c. Are natural proteins in the body that regulate wound and tissue regeneration.
   d. Stimulate angiogenesis by deflecting preosteoblasts during the treatment of periodontal defects.

3. The optimum amount of bone morphogenic protein suggested to enhance bone growth is approximately:
   a. 1 µg/ 40 mg weight of explant.
   b. 2 µg/40 mg weight of explant.
   c. 10 µg/40 mg weight of explant.
   d. 20 µg/40 mg weight of explant.

4. Allografts are typically freeze-dried and treated to prevent disease transmission. Frozen iliac cancellous bone and marrow, freeze-dried bone, and demineralized freeze-dried allografts can be obtained from commercial tissue banks.
   a. Both statements are true.
   b. Both statements are false.
   c. The first statement is true, the second statement is false.
   d. The first statement is false, the second statement is true.

5. Which growth and differentiation factors have been identified as potential contributors for periodontal regeneration?
   a. Insulin-like growth factor.
   b. Platelet-derived and fibroblast growth factors.
   c. Transforming growth factor-beta and bone morphogenetic proteins.
   d. All of the above.

6. All of the following statements are true EXCEPT:
   a. Insulin-like growth factor-1 may stimulate mitogenic activity in periodontal ligament cells.
   b. Insulin-like growth factor-1 better stimulates bone formation alone than in combination with PDGF.
   c. Insulin-like growth factors are produced primarily by the liver and circulate in the vascular system.
   d. Fibroblast growth factors are mitogenic and chemotactic for fibroblasts, chondrocytes, osteoblasts, and endothelial cells.

7. What is the average bone fill required during the use of autogenous bone grafts for treatment of many intraosseous defects?
   a. 1.5 mm.
   b. 2.5 mm.
   c. 3.5 mm.
   d. 4.5 mm.

8. Which healing process has been associated with the use of allografts?
   a. Osteoinduction.
   b. Osteoconduction.
   c. Both a and b.
   d. Neither a nor b.

9. Which factor may negatively influence the bioactivity of the DFDBA preparation?
   a. Various processing factors.
   b. Improper storage conditions.
   c. Delaying the procurement of bone from a donor.
   d. All of the above.

10. Since particular growth factors work only to stimulate specific cell types, a combination of GDFs that stimulate wound closure and the formation of hard and soft tissues is necessary. This statement is:
    a. True.
    b. False.